

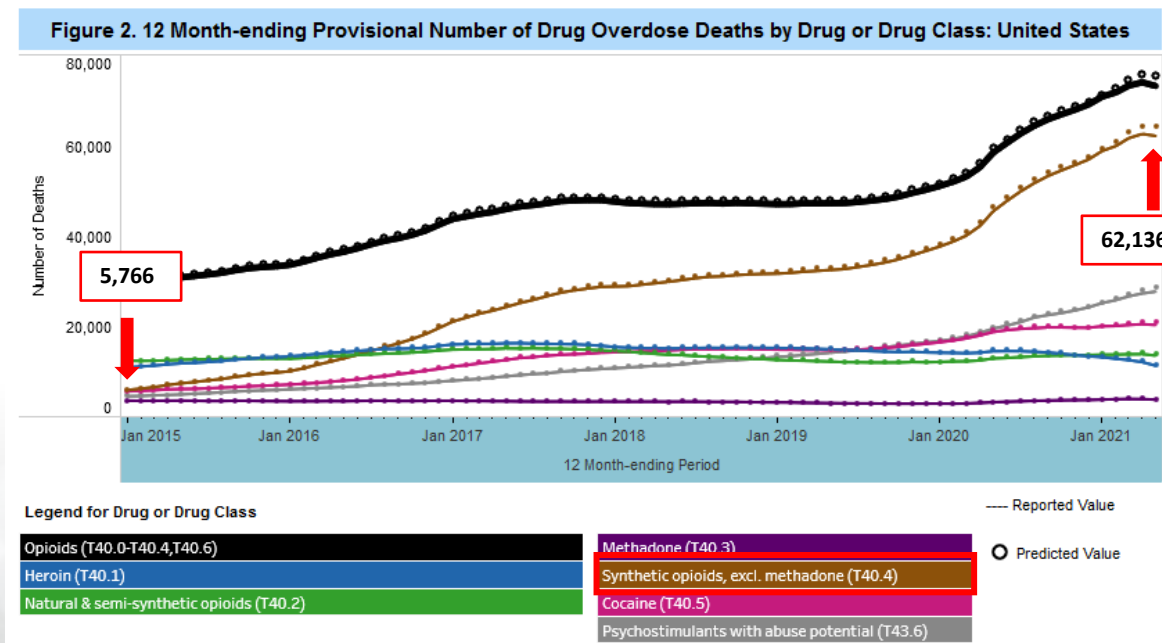
# Fentanyl Analogs & Harms Reduction



# Overview



- Opioids are now one of the leading causes of death among US adults
- Deaths from synthetic opioids has risen from 5,766 in January 2015 to 62,136 in May 2021
- Comparatively, 37,595 deaths from motor vehicles were reported in 2019
- Main culprit in these overdoses is fentanyl



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DEA Seized Enough Fentanyl to 'Kill Every Single American' This Year as Opioid Deaths Top 100K

**Drug overdoses driving down U.S. life expectancy – health officials**

**NEWS**  
**Texas seized enough fentanyl to kill 200 million people this year alone, officials say**

**NATIONAL TRAGEDY Fentanyl becomes 'leading cause of death for Americans aged 18 to 45' with more fatalities than COVID, cancer & suicide**

**Fentanyl overdoses become No. 1 cause of death among US adults, ages 18-45: 'A national emergency'**  
More adults between 18 and 45 died of fentanyl overdoses in 2020 than COVID-19, motor vehicle accidents, cancer and suicide

**'It's devastating': how fentanyl is unfolding as one of America's greatest tragedies**  
More than 100,000 people died from overdoses in a single year - driven primarily by one drug

- A study by Johns Hopkins found that most drug users were interested in **checking their drugs for fentanyl**
- 86% of participants showed interest in **knowing the amount of fentanyl** in their drugs
- 70% of participants reported that knowing that their drugs contained fentanyl would **cause them to change their behavior**
- Most drug users **prefer to not consume fentanyl** and **do not want to risk a lethal overdose**

### Why is this important?

- Many drug users buy drugs without knowing that they may be laced with fentanyl
- Even drug traffickers are often unaware of the drug's true contents



- Fentanyl tests can be used on drug powders to check whether the drugs contain fentanyl
- Being aware of fentanyl in their drugs will allow drug users to make an informed decision about whether to take the drug or how much to take
- This will not stop drug users from doing drugs, but it can prevent many accidental overdoses, leading to fewer deaths



A close-up photograph of a glass mortar and pestle. The mortar is on the left, and the pestle is on the right. Both are filled with white, round pills. The background is a soft, out-of-focus blue. The text "Fentanyl Analogs" is overlaid in the center in a white, sans-serif font.

# Fentanyl Analogs

## What is fentanyl?

- Highly addictive synthetic opioid
- 50-300x more potent than morphine
- Originally developed for use as an analgesic (pain reliever) and anesthetic
- Can be taken orally (e.g., pill, tablet), smoked, snorted, or absorbed through a transdermal patch
- Induces a high similar to effects of other opioids, such as morphine
- Often added to other drugs to increase their potency
- Associated with more than half of drug overdose deaths and rising



Fentanyl Consumed	Risk of Overdose
0.00005 grams	Death unlikely
0.0001 grams	Moderate risk of death
0.00015 grams	Significant risk of death
0.00025 grams	High risk of death
0.00004 grams	Very high risk of death
0.0007 grams	Death likely
0.001 grams	Near-certain death
0.002 grams	Death certain

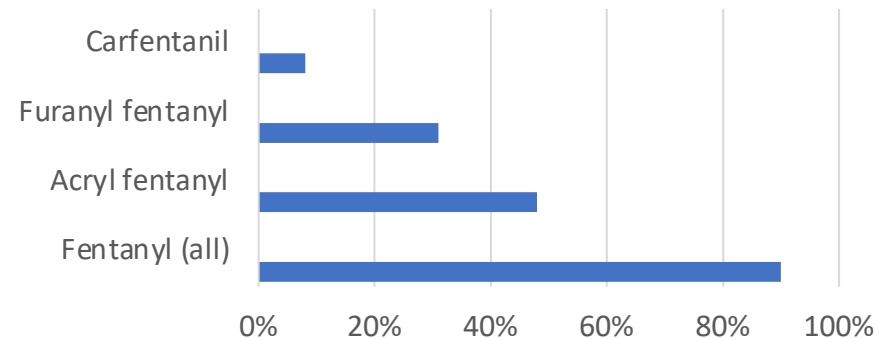
## Wright State University and MCCO/MVRCL identified drugs associated with overdose fatalities in Ohio during January-February 2017

- 90% of deceased tested positive for fentanyl
- 48% for acryl fentanyl
- 31% for furanyl fentanyl
- 8% for carfentanil
- Most tested positive for multiple types of fentanyl

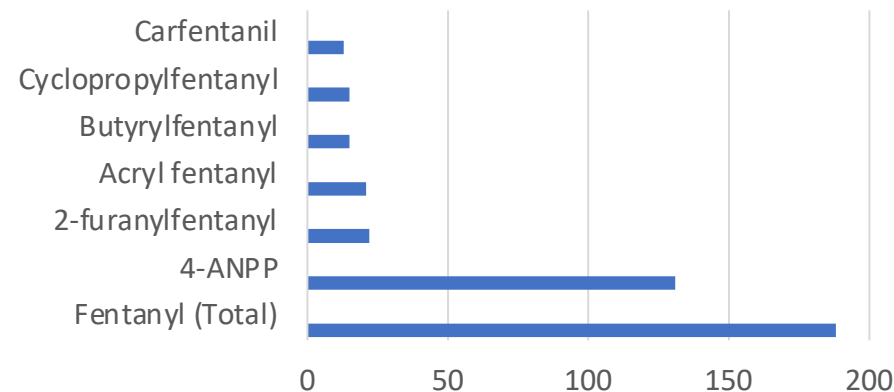
## Chhabra et al. screened urine samples from living patients that were positive for fentanyl or opiates

- 65.3% samples contained at least one analog
- 26.0% were positive for multiple analogs

Fentanyl & Fentanyl Analog Positivity in Overdose Fatalities



Fentanyl & Fentanyl Analog Positivity in Living Patients

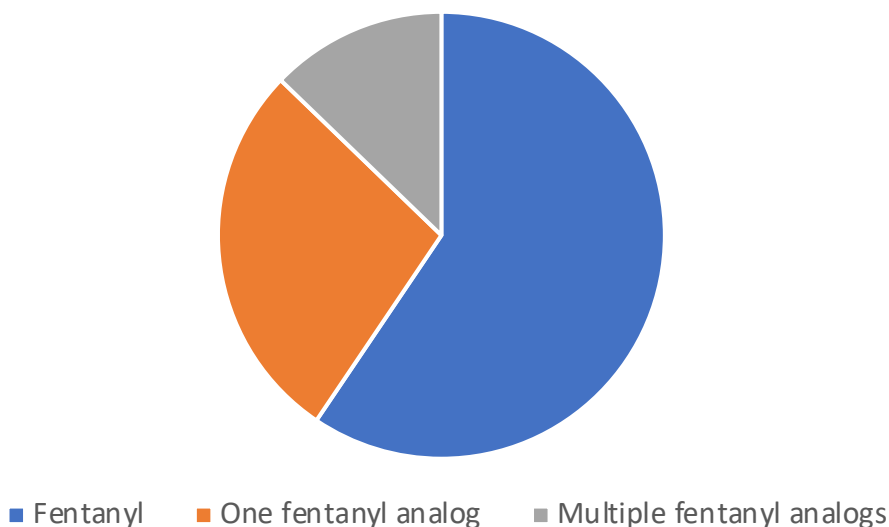




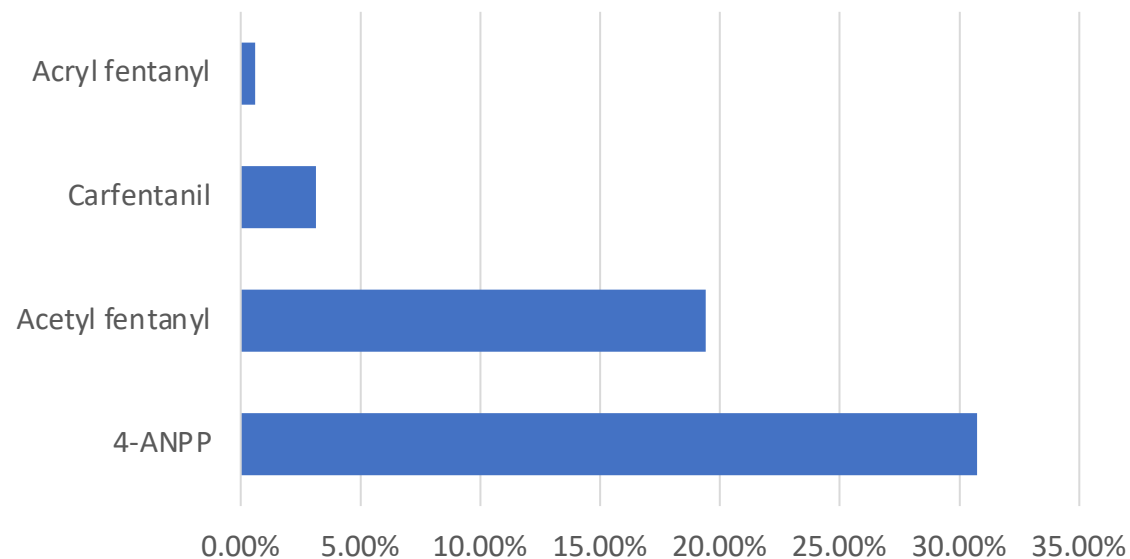
## Millennium Health analyzed urine samples collected between July 2019-March 2020 from across the US

- In patients testing positive for fentanyl without a fentanyl prescription, 40.55% were positive for fentanyl analog and 12.78% were positive for multiple analogs
- Analog positivity rate was 8.93% among those who were prescribed fentanyl

Fentanyl Analog Positivity Rates in Non-Prescribed Population



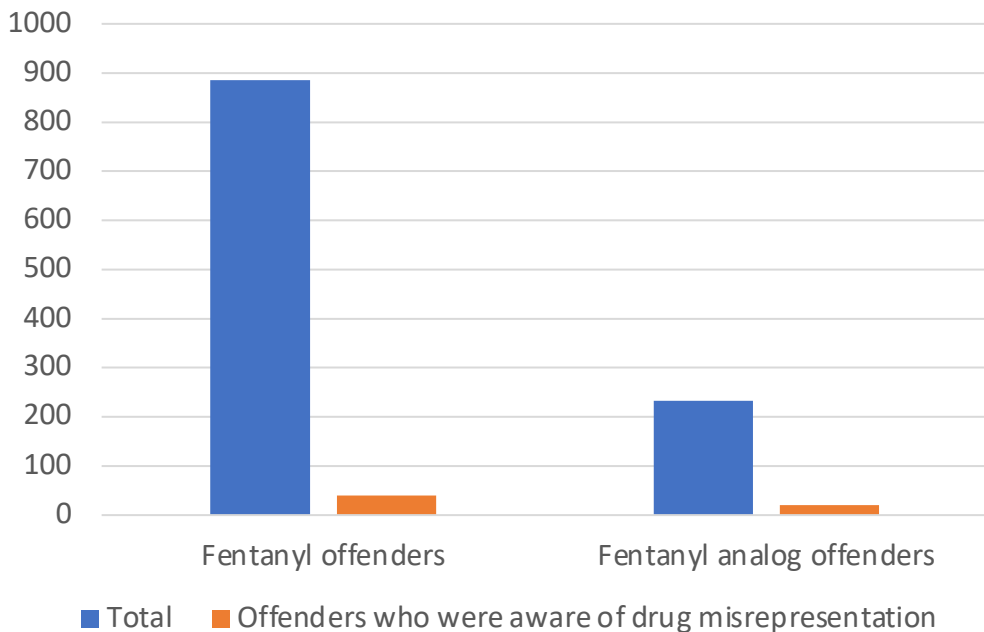
Top 4 Fentanyl Analogs Found in Non-Prescribed Population



## United States Sentencing Commission investigated federal cases of drug trafficking in 2019

- Number of fentanyl offenders rose from 25 (in 2015) to 886
- Number of fentanyl analog offenders rose from 4 (in 2016) to 233
- Only 4.5% of fentanyl offenders and 9.0% of fentanyl analog offenders knowingly sold these substances as a different drug

United States Sentencing Commission



Fentanyl Analogues  
N=233

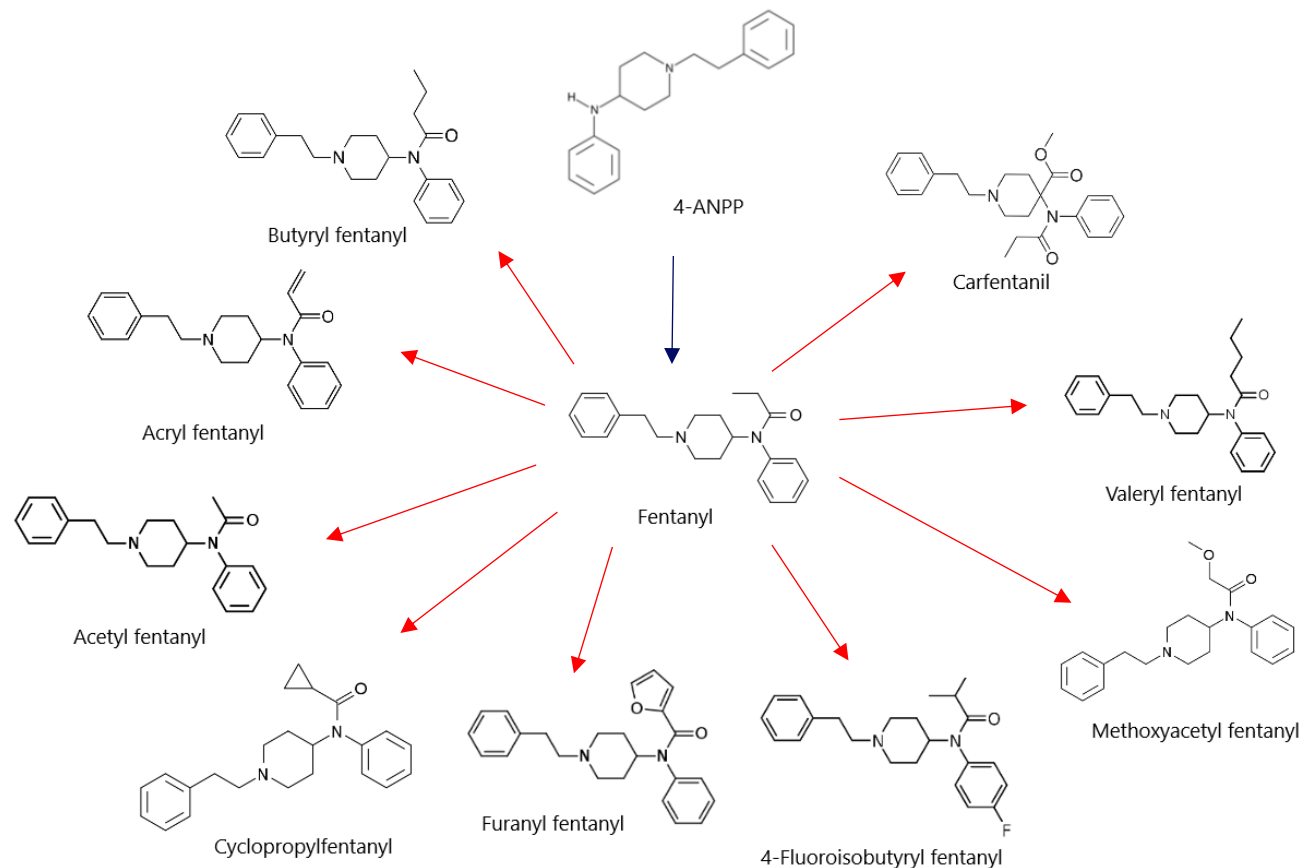
Primary Analogue	%	N
Carfentanil	27.5%	64
Furanyl Fentanyl	19.3%	45
Acetyl Fentanyl	12.5%	29
4-Fluoroisobutyryl (Para-Fluoroisobutyryl) Fentanyl	9.0%	21
Cyclopropyl Fentanyl	8.2%	19
Butyryl Fentanyl	6.0%	14
Methoxyacetyl Fentanyl	4.7%	11
Acryl Fentanyl	3.9%	9
Other <sup>130</sup>	9.0%	21

## Fentanyl analogs

- Analogs are variations of the original drug that have slight differences in their chemical structure
- They are becoming increasingly more common on the illicit market and contributing to more overdose-related deaths

### Common analogs include:

- Carfentanil (10,000x more potent than morphine)
- Furanyl fentanyl
- Acryl fentanyl
- Acetyl fentanyl
- 4-fluoroisobutyryl fentanyl

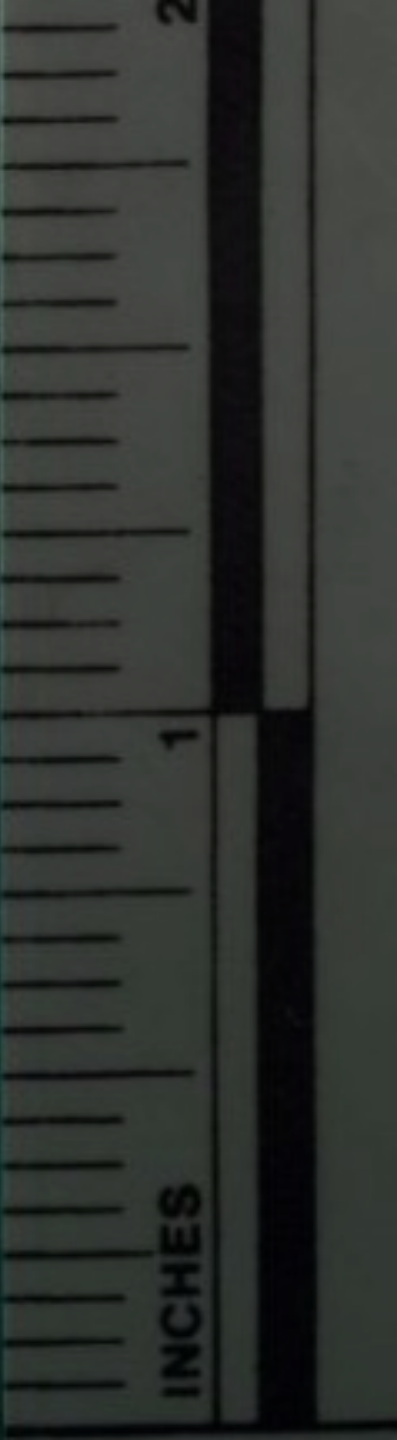


# Analog Detection

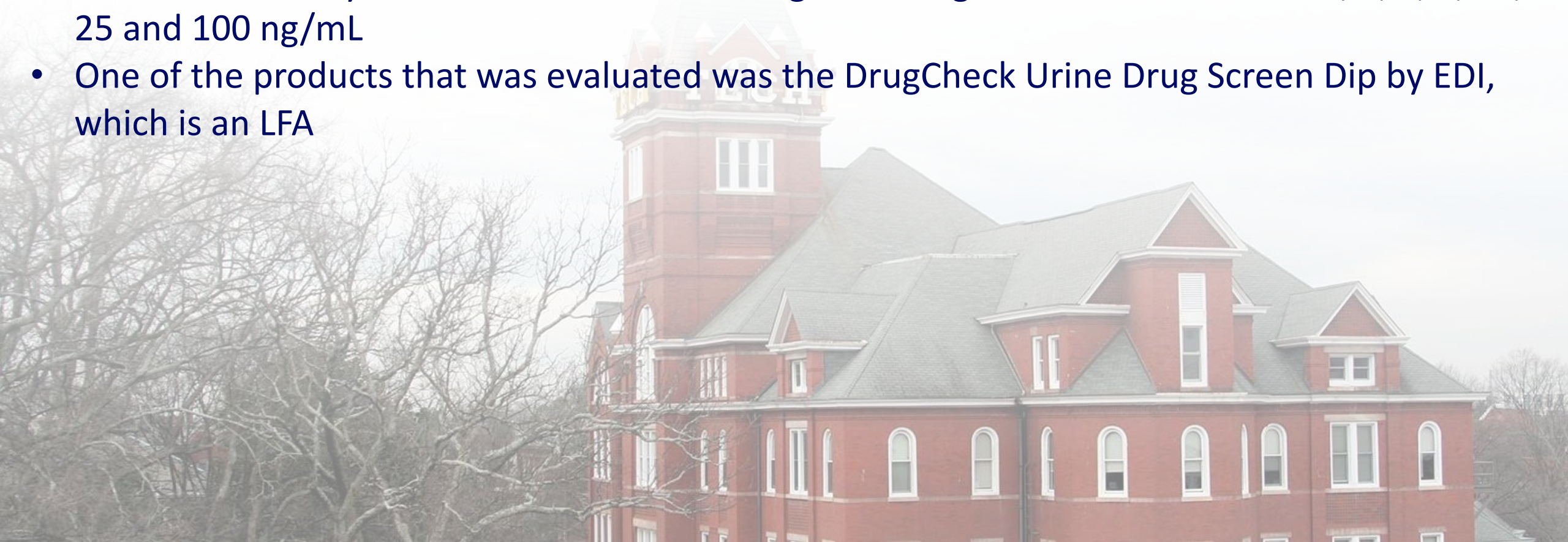
CARFENTANIL

FENTANYL

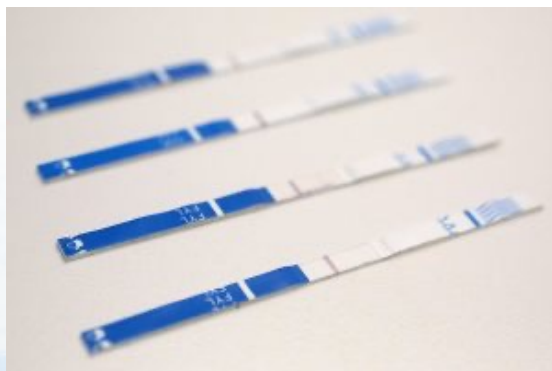
HEROIN



- Wharton et al. conducted a study demonstrating the effectiveness of commercial immunoassays in detecting fentanyl analogs
- Immunoassays used in the study include LFAs, ELISAs, and EMITs targeting fentanyl, norfentanyl, and carfentanil
- The immunoassays were each evaluated using 30 analogs at concentrations of 1, 2, 3, 5, 10, 25 and 100 ng/mL
- One of the products that was evaluated was the DrugCheck Urine Drug Screen Dip by EDI, which is an LFA



## Lateral flow assay (LFA)



- Fastest turnaround time (~5 min) and inexpensive
- No laboratory instruments required
- Sample is added to the test device and migrates up the test strip
- Antibodies on the test strip react with the sample solution to produce results

## Enzyme-linked immunosorbent assay (ELISA)



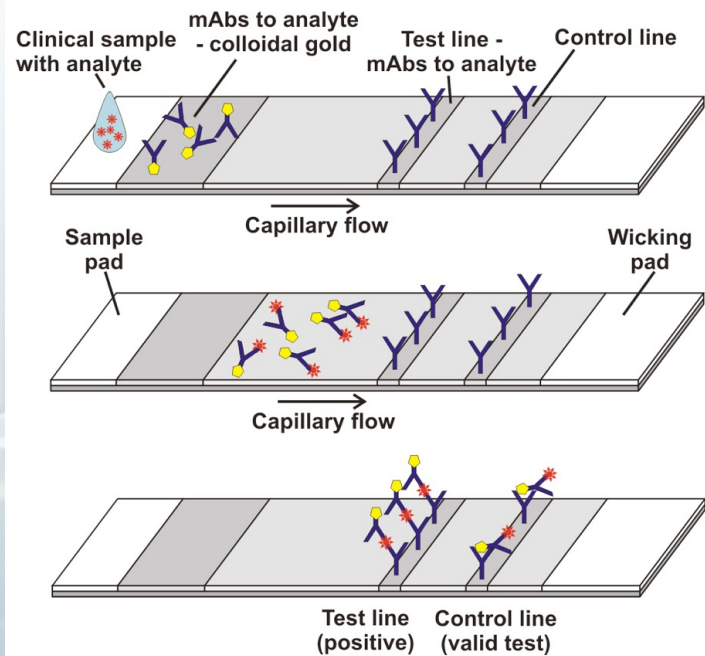
- Most sensitive, but slow and expensive
- Laboratory instruments required
- Capture antibody, sample, and detection antibody are added to a plate
- A substrate is added that causes a color-forming reaction
- Color is read by plate reader to determine results

## Enzyme multiplied immunoassay technique (EMIT)

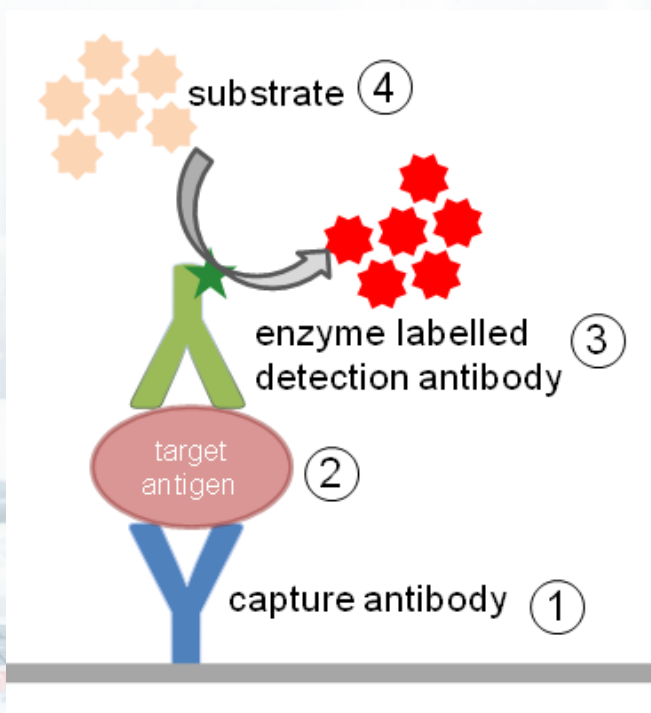


- Relatively quick turnaround time (~30-60 min) but expensive
- Laboratory instruments required
- Enzyme-linked drug is added to sample and competes with drugs for antibody binding
- Free enzymes cause a change in color that is measured by spectroscopy to determine results

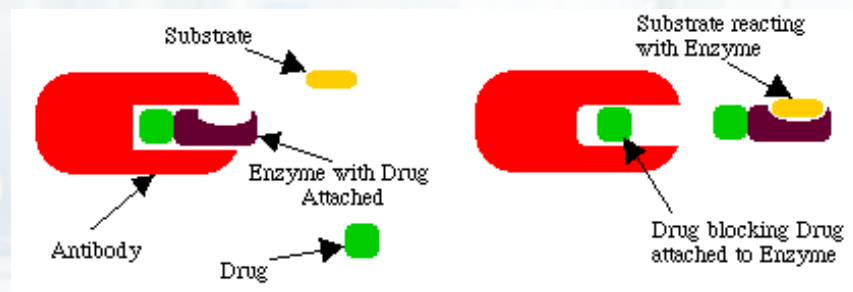
### Lateral flow assay (LFA)

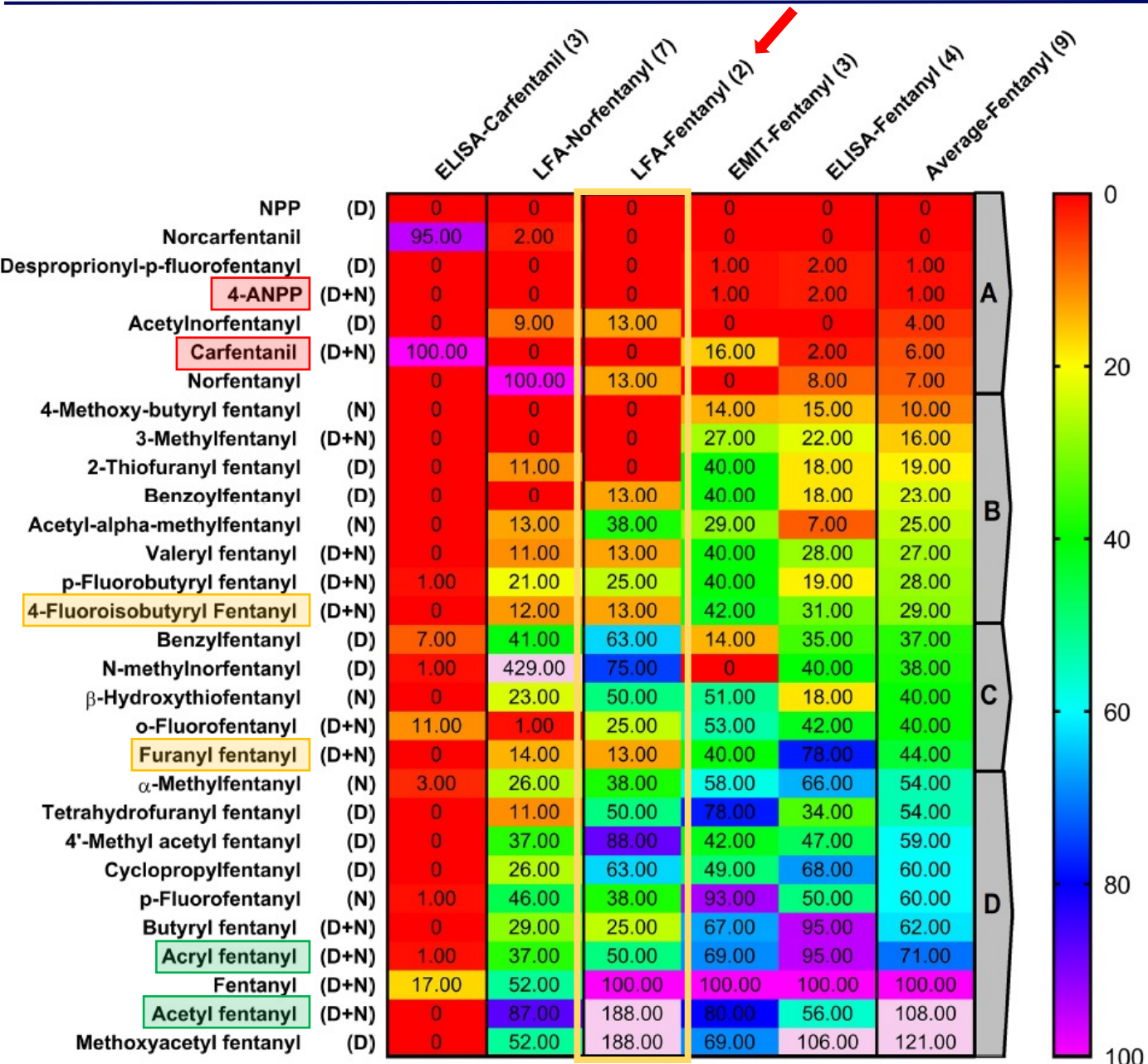


### Enzyme-linked immunosorbent assay (ELISA)

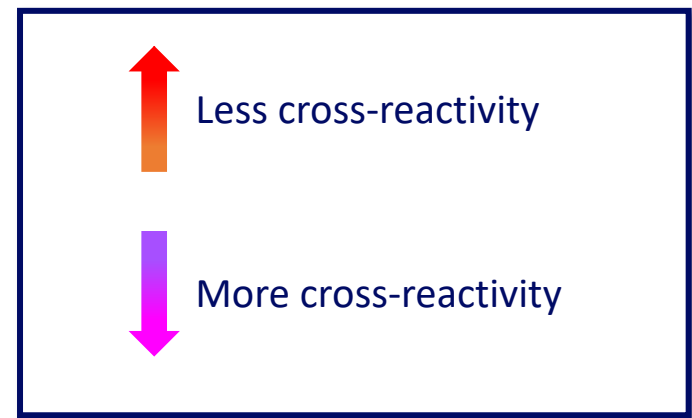


### Enzyme multiplied immunoassay technique (EMIT)





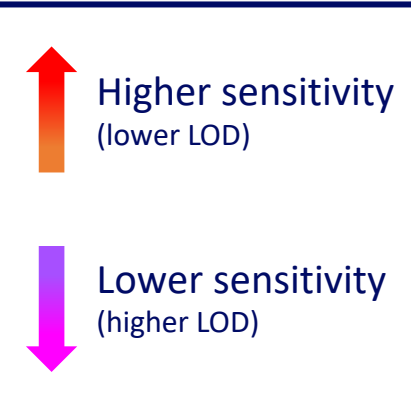
Average percent cross-reactivity of commercial immunoassays against 30 fentanyl analogs, separated by assay type and target drug





Kit Reported Cutoff (ng/mL)	BTNX		PreScreen Plus						DrugCheck
	AlcoPro	CLIA waived	NarcoCheck	Medimpex	Quickscreen	Speares Medical			
	20	10	200	25	10	20	75	20	10
Fentanyl	100	100	200		20	80	400	400	40
Norfentanyl		400	20	40	20	80	200	20	40
Acetyl Fentanyl	40	80	200		10	40			20
Acetyl- $\alpha$ -methylfentanyl	400	200			40	400			200
Acetylnorfentanyl		400	200	400	200			100	400
Acryl Fentanyl	200	200	200		40	80			40
$\alpha$ -Methylfentanyl	200	400	400		20	200			100
4-ANPP									
Benzoylfentanyl (Phenyl fentanyl)	400								
Benzylfentanyl	400	100	400		20	100			40
$\beta$ -Hydroxythiofentanyl	200	200	400		80	100			80
Butyryl Fentanyl	200		400		40	80			80
Carfentanil									
Cyclopropylfentanyl	100	400	400		40	100			80
Despropionyl-p-fluorofentanyl									
4-Fluoroisobutyryl Fentanyl	400				80	200			200
Furanyl Fentanyl	400				80	400			80
Methoxyacetyl Fentanyl	40	80	200		20	80	400	400	40
4-Methoxy-butyryl Fentanyl									
4'-Methyl Acetyl Fentanyl	80	200	200		40	80			40
Norcarfentanil									
3-Methylfentanyl						400			400
N-Methylnorfentanyl	400	80	10	10	5	20	20	10	10
N-Phenethyl-4-piperidinone									
o-Fluorofentanyl	200					400			
p-Fluorofentanyl	200	400	100			20	80		40
p-Fluorobutyryl Fentanyl	200		400			40	200		80
Tetrahydrofuranyl Fentanyl	100					100	400		100
2-Thiofuranyl Fentanyl							200		100
Valeryl Fentanyl	400						400		400

- WHPM’s DrugCheck fentanyl test was used and was able to detect 22 of the 30 analogs included in the study
- WHPM’s DrugCheck outperforms most of the competitor tests used in the study
- Table on right shows the lowest concentration (in ng/mL) at which the analog listed in each row was detected



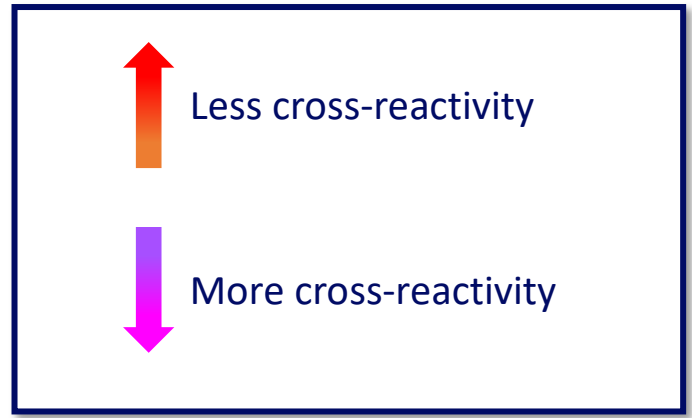
Higher sensitivity  
(lower LOD)

Lower sensitivity  
(higher LOD)

**Note:** The color scale in this table is the opposite of what was in the previous table. Here, red = high sensitivity (low LOD) and purple = low sensitivity (high LOD)

## Fentanyl analog detection

- LFAs are a low-cost solution that makes drug testing widely available and accessible
- Many competitor LFAs are only validated for detection of one fentanyl analog or metabolite



Fentanyl Analog	Cross-Reactivity	Fentanyl Analog	Cross-Reactivity	Fentanyl Analog	Cross-Reactivity
NPP	0%	Benzoylfentanyl	13%	α-Methylfentanyl	38%
Norcarfentanil	0%	Acetyl-alpha-methylfentanyl	38%	Tetrahydrofuranyl fentanyl	50%
Despropionyl-p-fluorofentanyl	0%	Valeryl fentanyl	13%	4'-Methyl acetyl fentanyl	88%
4-ANPP	0%	p-Fluorobutyryl fentanyl	25%	Cyclopropylfentanyl	63%
Acetylnorfentanyl	13%	4-Fluoroisobutyryl fentanyl	13%	p-Fluorofentanyl	38%
Carfentanil	0%	Benzylfentanyl	63%	Butyryl fentanyl	25%
Norfentanyl	13%	N-methylnorfentanyl	75%	Acryl fentanyl	50%
4-Methoxy-butyryl fentanyl	0%	β-Hydroxythiofentanyl	50%	Fentanyl	100%
3-Methylfentanyl	0%	o-Fluorofentanyl	25%	Acetyl fentanyl	188%
2-Thiofuranyl fentanyl	0%	Furanyl fentanyl	13%	Methoxyacetyl fentanyl	188%

**Above:** Average cross-reactivities of fentanyl analogs among commercial LFAs targeting fentanyl

- LFAs targeting fentanyl or norfentanyl can detect many (but not all) fentanyl analogs
- Some manufacturers opt to make a separate test targeting carfentanil as it is a common analog that both fentanyl and norfentanyl LFAs consistently fail to detect

